



Lymph Notes

*“ it’s better ..
Together “*

Biochemistry First year

2nd term

Success is no accident; it is hard work, perseverance, sacrifice, hours of studying and most of all.. it is the love and passion to what you do and managing to learn then practice it well.

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ENZYMES

1. Give an account on Isoenzymes:- Meaning and chemical significance (2011) , characteristics and examples (2012), one example in detail (2015 September) .

They are group of enzymes characterized by :-

- catalyze same reaction
- have different affinity polypeptide chains produced by different genes
- separated by electrophoresis (different migration rate)
- have different affinity to substrate
- affected differently by the different activators and inhibitors
- present in same or different cells Examples

:-

***Lactate dehydrogenases (LDH)**

- Tetrameric enzyme
- Of four protomers (subunits) of two units H&M
- Tetrameric molecule is the only active form of enzyme
- Different subunits combined to form 5 Isozymes → $I_1 (H_4)$, $I_2 (H_3M_1)$, $I_3 (H_2M_2)$, $I_4 (H_1M_3)$, $I_5 (M_4)$ → H&M subunits are synthesized by distinct genes and differentially expressed in different tissues
- Estimation of LDH in plasma is of clinical importance
- Isozyme 1(H_4) → is mainly of cardiac origin and its level in plasma increases in case of myocardial infarction
- Isozyme 5(M_4) → increases in plasma in cases of liver diseases (hepatic origin) or muscle disease (muscular origin)

***Creatine kinase (CK) or creatine phosphokinase (CPK) →**

Dimer formed of two subunits B&M and has three Isozymes :-

- CK1 or (CK-BB) :- in brain tissues, its level in plasma increases in brain infarction
- CK2 or (CK-BM) :- present in cardiac muscle more than in skeletal muscles, so its plasma level increases markedly in myocardial infarction
- CK3 or (CK-MM) :- present in skeletal muscle more than in cardiac muscles, so its plasma level increases markedly in muscle diseases

2. Give an account on Competitive and allosteric inhibitors .

Competitive inhibitors *Competitive

inhibition occurs at active site .

*Competitive inhibitor is structurally similar to that of substrate , *So it compete with substrate to bind at active site .

*Inhibition occurs when it binds at the active site and is reversible .

No effect on V_{max} :-

*Their effect is reversed by increasing $[S]$ (substrate concentration) .

*At a sufficiently high substrate concentration, the reaction velocity reaches V_{max} observed in absence of inhibitor .

Increase of K_m :-

*Competitive inhibitor increases the K_m for a given substrate .

* i.e :- more substrate is needed to achieve $\frac{1}{2} V_{max}$.

Examples :-

1- Malonate

→ Inhibits succinate dehydrogenases enzyme as they are structurally similar .

2- Sulfonamides

→ Used in treatment of bacterial infections .

→ Bacteria synthesize folic acid from p aminobenzoic acid (PABA) .

→ Sulfonamide drugs contain sulfanilamide a structural analog of PABA .

→ When used as chemotherapeutic agent, it blocks synthesis of folic acid in bacteria which is essential for bacterial multiplication .

→ Act as bacteriostatic agent .

→ Competitive inhibitor for enzyme involved in formation of folic acid using PABA as substrate .

3- Allopurinol

→ Treatment of gout .

→ Gout due to excessive production of uric acid .

→ Xanthine oxidase involved in formation of uric acid from hypoxanthine . → Allopurinol is structural analog of hypoxanthine, so it is anti metabolite of hypoxanthine .

→ Blocks formation of uric acid by inhibiting xanthine oxidase .

4- Dicumarol & warfarin

→ Anticoagulants .

→ Structurally similar to vitamin K (required for activation of clotting factors) .

Allosteric inhibitors

*Small organic molecule binds to specific site away from catalytic site .

*Produce conformational changes in protein structure that lead to decrease activity of enzyme .

*e.g. ATP for phosphofructokinase-1

*Decrease affinity of enzyme to its substrate (Increase K_m) or,

*Decrease maximal catalytic activity (Decrease V_{max}) or both .

→ Allosteric modifiers include both allosteric activators and allosteric inhibitors .

Feedback inhibition :-

→ Inhibition of the activity of enzyme in a pathway by the end products of this pathway .

→ May occurs through binding of end product with allosteric site present on regulatory key enzyme

→ This is usually occurs at the earliest irreversible step unique to that particular pathway.

→ This prevents accumulation of unwanted amount of metabolic end products . →

In case of branched biosynthetic pathways, multiple feedback loops are more common for regulation of enzymes of these pathways .

Cumulative feedback inhibition : inhibition of one enzyme by two or more products

3. Mention two differences between competitive and allosteric INHIBITORS .

Competitive Inhibitors	Allosteric inhibitors
- Reversible inhibitors . - The inhibitor is similar (structural analogue) to the substrate . - The inhibitor competes the substrate for the active site of the enzyme . - K_m : Increased V_{max} : No effect - Examples :- 1- Allopurinol 2- Warfarin & dicumarol	- IRReversible inhibitors . - Are organic molecules that binds to the allosteric site → conformational changes in the active site → less suitable to bind substrate - The end product of a series of reactions inhibits an enzyme early . - K_m : Increased V_{max} : Decreased

FREE NUCLEOTIDES

1. Enumerate

a) Two cyclic nucleotides and two nucleotides coenzymes (2007)?

□ **Cyclic nucleotides:** * C-AMP (Cyclic 3',5' Adenosine monophosphate).

C-GMP (Cyclic 3',5' Guanosine monophosphate).

□ **Nucleotides coenzymes:**

*NAD⁺ (Nicotinamide Adenosine Dinucleotide)

*NADP (Nicotinamide Adenosine Dinucleotide Phosphate).

○ *coenzyme A.

B) Two adenine containing nucleotides : one which acts as a second hormone messenger and one which acts as a sulfate donor:

2nd hormone messenger: CAMP (Cyclic 3',5' Adenosine monophosphate).
sulfate donor: PAPS (3'phosphoadenosine 5' phosphosulfate).

c) Two minor bases: one purine and one pyrimidine present in tRNA .(2000)

-purine : N⁶ methyladenine. \ N⁶N⁶ dimethyladenine \ 7-hydroxymethylsytosine
-pyrimidine: dihydrouracil.

D) Four pyrimidine bases found in nucleic acids.(2009)

-cytosine (in DNA & RNA). -thymine (.minor base in RNA & mainly DNA)
-5 methylecytosine. -5 hydroxymethylecytosine.
-Dihydrouracil. -uracil (in RNA)

e) Four adenine containing nucleotides.

-ADP (Adenosine diphosphate) -AMP (Adenosine monophosphate)
-ATP (Adenosine triphosphate) -SAM (S-adenosyl methionine)
-PAPS (3'phosphoadenosine 5' phosphosulfate)
-NAD⁺ (Nicotinamide Adenosine Dinucleotide)
-NADP⁺ (Nicotinamide Adenosine Dinucleotide Phosphate) -
Coenzyme A.

2. What are the hydrolytic products of :

- **SAM : (2009)**

Adenine + ribose + methionine. •

Inosonic acid : (2008)(2009)

Hypoxanthine + ribose + phosphate.

• **ATP:**

Adenine + ribose + 3 phosphates.

3. Name and importance of a free nucleoside and a free nucleotide containing pantothenic acid. (2011)

-free nucleoside : -SAM→ methyl donor.

-free nucleotide containing pantothenic → coenzyme A→ acetyl group carrier .

4. Explain on biochemical basis:

*** Role of G-proteins as signal transducers (2004,2011,2014) ***

Structure & mechanism of action (sep 2015) - Structure:

* G-proteins are trans membrane proteins formed of 3 subunits (α , β , γ) and interact with cell membrane receptor in presence of ligand.

* There are many isoforms of G-proteins which produce different effects on same cell or different cells. The α subunit is the guanylate nucleotide binding unit.

- Mechanism:

- Binding of ligand to its cell membrane receptor produces conformational changes that lead to binding of the receptors to G-proteins.
 - This step activates G-proteins and replacement of GDP with GTP at the binding site of α subunit which is released bounded to GTP.
 - The released α subunit-GTP complex produces its effect on certain effector proteins which results in generation of second messenger and mediation of cellular response (stimulation or inhibition).
 - GTP is then hydrolyzed to GDP and PI (α subunit acts as GTPase) and in the same time α subunit rebinds with β & γ subunits to form intact G-protein. •
The cycle may be repeated several times as long as the ligand is present.
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5. Role of CAMP as second messenger (2005,2015).

- Hormone-receptor interaction activates G-protein which activates adenylyl cyclase that forms CAMP from ATP.
 - Cyclic AMP activates protein kinase A which causes phosphorylation of proteins (eg:Enzymes) causes either its activation eg: glycogen phospholylase kinase or its inactivation eg: glycogen synthase.
 - This process is reversed by protein phosphatase which dephospholyrates enzymes.
 - Many hormones act through this mechanism eg: glycogen & epinephrine activate adenylyl cyclase and increase CAMP (inhibits protein phosphatase).
 - Insulin activates protein phosphatase and phosphodiesterase and decreases CAMP level so it reverses effects of glycogen & epinephrine.
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NUCLEICACID

1. Transfer RNA (structure & Function)

Structure : contain unusual bases (**dihydrouracil**) and have extensive intra-chain base-pairing that lead to characteristic secondary and tertiary structure (**clover leaf appearance**) .

It has five main arms :-

1- The acceptor arm : of tRNA terminates at its 3' OH by a specific sequence formed of CCA . Amino acids are carried in the form of aminoacyl group connected to the 3' hydroxyl group of the acceptor arm .

2- D arm : contains the unusual base dihydrouracil . D arm is important in recognition of tRNA by aminoacyl - tRNA synthetase enzyme which catalyzes binding of a specific AA to its specific tRNA.

3- Anticodon arm : contain three specific bases known as the anticodon . This portion of tRNA plays a key role in translation by pairing with the complementary codon of mRNA and helps in placing each amino acid in its proper site in the polypeptide chain of protein .

4- Extra arm : contain from 3 to 12 bases and it's the major site for variation in tRNA .

5- Loop IV : contain the unusual specific sequence of **thymine and pseudouridine** bases . This arm is important for binding of tRNA to the ribosome during the process of protein synthesis .

Its main Function :

The tRNA serve as an adapter molecule that carries its specific amino acid to site of protein synthesis . It recognize the genetic code word of mRNA, which specifies the addition of its amino acid to the growing peptide chain .

2. DNA and RNA (Four differences)

	DNA	RNA
SUGAR	Deoxyribose .	Ribose .
TYPES	Circular or linear (A,B,Z forms)	mRNA , tRNA , rRNA
FUNCTION	Genetic information and synthesis of RNAs .	Protein synthesis
SITE	Nucleus and Mitochondria .	Mainly in cytosol . Less commonly in nucleus and mitochondria .
SHAPE OF STRAND	Double helix	Single strand

NITROGENEOUS BASES	Adenine, guanine, cytosine, thymine (no uracil)	Adenine, guanine, cytosine, uracil (thymine as minor base in tRNA)
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3. Mention two minor bases :- one purine and one pyrimidine present in tRNA .

**Purine → Dihydrouracil pyrimidine
→ Thymine .**

4. Explain in details the primary and secondary structure of B form of DNA.

Primary structure :-

- There are 4 main nucleotides that form DNA (dAMP, dGMP, dCMP, dTMP) . - In each strand, the nucleotides are linked together by phosphodiester bonds between the 5`hydroxyl of one nucleotide and 3`hydroxyl of the next nucleotide .
- The alternating sugar phosphate units form the backbone of each DNA strand (5`-ps-p-s-3`) .
- The nitrogenous bases, which are linked to the pentose, are projecting to the inside of the 2 strands of DNA at right angle .
- The sequence of bases determines the coding structure of DNA .
- Each polynucleotide has 2 terminals :
 - * One end has free phosphate group attached to 5`- hydroxyl group of the terminal pentose .
 - * The other end has a free 3`- hydroxyl group .
- The order of nucleotides in any DNA strand is always written in the 5` to 3` direction .

Secondary structure :-

Watson and crick :- Proposed a structure for DNA in the form of a double helix and it's now referred as

B- form of DNA . It has the following characters :- **1- Two**

antiparallel strands form a right handed helix :-

The 2 strands of DNA are paired to each other and coil around a common axis to form a right handed helix . The 2 strands run antiparallel. One runs in the 3` → 5` direction and the other runs in the 5` → 3` direction .

2- Complementary base pairing :-

The 2 strands are held together by the complementary base pairing through specific hydrogen bonds

- Adenine pairs with thymine through two hydrogen bonds .
- Guanine pairs with cytosine through 3 hydrogen bonds .

Therefore the number of adenine bases equal the number of thymine bases and the number of cytosine bases equal the number of guanine bases .

The sequence of the 2 strands is complementary and the sequence of one strand determines the sequence of the other one . And this is important in DNA replication .

3- base stacking :-

The base pairs inside the helix are stacking above each other by van der Waals forces and hydrophobic interaction .

The hydrogen bonding between complementary base pairs , van der Waals forces and hydrophobic interactions of stacked base pairs provide the stability of the double helix .

4- Spiral staircase :-

The double helix of DNA appears much like spiral staircase, in which there are 10.4 base pairs or steps for each complete turn of the helix . Each complete turn is 3.4 nm long .

5- dimensions:-

The B- form of DNA is 2 nm wide .

From outside of the helix, two grooves are apparent, a major groove (2.2 nm) and a minor groove (1.2 nm) .

Through these grooves many drugs and proteins can make contact with the nitrogenous bases without any need to open the helix .

DNA¹¹

1. The role of DNA-dependant-RNA-Polymerase and RNA-dependantDNA-Polymerase in replication.

- DNA dependant RNA Polymerase: initiation of DNA synthesis by formation of RNA primers. At replication fork, both strands of DNA serves as a templates for DNA synthesis. To initiate DNA synthesis , RNA primers is formed by the action of DNA-dependant-RNA-Polymerase are elongated by DNA Polymerase α

- RNA-dependant-DNA-Polymerase : reverse transcription as function of telomerase:

Special kind of reverse transcriptase. It is made of protein part and RNA strand.

It uses its RNA strand as a template for synthesis of complementary DNA strand.

In that RNA are copies of A/C sequence that is complementary to the T/G repeat sequence. The RNA base pair with the terminal nucleotides at the singlestranded 3' end of the DNA strand.

Telomerase recognizes the single strand 3' terminus and uses its RNA molecule as a template to elongate the parental strand by 100 nucleotides and the process may be repeated. The elongated end is used as a template for completing synthesis of telomere of the lagging strand by DNA polymerase.

2. Causes and manifestation of xeroderma pigmetosum (2011)

- Causes : It is a genetic disease due to defective DNA repair.
- manifestation :It is a clinical syndrome characterized by hypersensitivity of sunlight and UV.
- resulting in increased skin cancer incidence and premature aging and death.

3. The importance of telomerase enzyme in replication. (2014, Sep 2015)

- There is a problem in replication of linear chromosomes while the leading strand can be synthesized to the very end the lagging strand cannot because:

- 1\ The primase cannot work to the very end of the template.
- 2\ Even in the likely event that a primase could start at the very end of the template, remove of RNA would have a short gap.

This problem is solved by telomerase which maintains the telomeric ends.

Telomerase is special kind of reverse transcriptase.

It is made of protein part and RNA strand. It uses its RNA strand as a template for synthesis of complementary DNA strand.

In that RNA are copies of A/C sequence that is complementary to the T/G repeat sequence. The RNA base pair with the terminal nucleotides at the singlestranded 3' end of the DNA strand.

Telomerase recognize the single strand 3' terminus and uses its RNA molecule as a template to elongate the parental strand by 100 nucleotides and the process may be repeated.

The elongated end is used as a template for completing synthesis of telomere of the lagging strand by DNA polymerase.

4. RNA editing changes mRNA after transcription. (2014)

- Coding information of mRNA can be changed by RNA editing for example:
 - In Liver : the single apolipoprotein gene (apoB-gene) is transcribed into mRNA that directs the synthesis of apo B-100 protein (100kDa)
 - In the intestine: the same gene directs the synthesis of primary transcript , then by the action of cystidine deaminase a CAA codon in mRNA is converted to UAA which is a termination codon apo B-48 protein (48kDa) is the result of translating this edited form of mRNA.
- Apo B-100 protein and Apo B-48 protein have different function.

5. The sigma factor and the Rho factor have different roles in prokaryotic RNA synthesis. (2010, 2014)

- Sigma factor :
 - Enables RNA polymerase to recognize the specific promotor regions of the DNA.
 - Increases the affinity of the holoenzyme to the promotor region.
- Rho factor :

Rho dependant termination: this process requires the participation of an additional protein, Rho factor binds to C-rich region near the 3' end of the newly synthesized RNA.

Rho factor has ATP-dependant-RNA-DNA helicase activity it hydrolyses ATP and uses energy to release the 3' end of the transcript from the template.

Explain on the biochemical basis:

1. Protection of 3' and 5' terminals of mRNA in Eukaryotes.

- Polyadenylation at 3' end: Poly (A) polymerase (PAP) is responsible for addition of poly (A) tail at the 3' end of pre-mRNA.

It stabilizes the mRNA and protects it against the attack by ribonucleases.

The length of poly (A) tail determines the half life time of mRNA.

It binds poly(A)tail-binding proteins which increases the efficiency of translation.

- Capping at 5' end :

Methyl-guanosine cap is added at 5' end through the following mechanisms:

1\ RNA triphosphatase hydrolyzes the terminal γ - phosphate of nascent pre-mRNA to form diphosphate.

2\ The Diphosphate is the capped with GMP by the enzyme RNA guanylyl transferase.

3\ GMP is methylated at position 7 by the use of S-adenosyl-methioine (SAM)

It protects 5' end from ribonucleases which are specific for 3'-5' phosphodiester bonds

2. Synthesis of antiparallel strands of DNA simultaneously through DNA polymerase can work in 5' -3' direction only (2010-2011)

- DNA –polymerase is only able to read the parental nucleotide sequence in the 3' to 5' direction (forms strand in direction 5' to 3') this can be completed by two different on each strand :
- Leading strand: it's copied in the direction of advancing replication fork and it is synthesized in continuous manner.
- Lagging strand: it is copied discontinuously in the opposite direction of the advancing replication fork in form of okazaki fragments. Similar to the leading strand, lagging strand is also copied in 5' to 3' direction.
- DNA –polymerase III has proof-reading activity. It hydrolytically removes the misplaced nucleotide (act as exonuclease) and replaces it with correct nucleotide. as misreading of template sequence may result in permanent mutations .

- (DNA –polymerase III has polymerase activity from 5' to 3' and exonuclease activity from 3' to 5')

1- Compare and Contrast:

1-RNA splicing and RNA editing. (Sep 2015)

Point of comparison	RNA splicing	RNA editing
Definition	Splicing removes introns (non expressed regions) and joins exons (expressed regions) for functional mRNA.	Coding information of mRNA can be changed by RNA editing.
Importance	• It decreases the incidence of mutations. • Alternative splicing may lead to the formation of different types of mRNA molecules or proteins.	• In the liver: The single apolipoprotein gene (apo B gene) is transcribed into an mRNA that directs the synthesis of apoB-100 protein (100 KDa) • In the intestine: The same gene directs the synthesis of a primary transcript, then by the action of cystidine deaminase, a "CAA" codon in mRNA is converted into "UAA" which is a termination codon, apoB48 protein (48 KDa) is the result of translating this edited form of mRNA.
Proteins and enzyme required	Splicing requires a spliceosome formed of special proteins and small nuclear RNA (snRNAs). snRNAs usually consist of about 100 nucleotides. The base composition of snRNA is complementary to the ends of introns. This helps snRNA and introns to base pair and this leads to loop formation. → leads to removal of intron loop and joining of exons takes place.	In intestine: editing requires cystidine deaminase to convert a "CAA" codon to "UAA" codon which is a termination codon.

Describe five of the following processes:

1. 1-Post transcriptional intron excision and splicing- give a disease condition related to the process. (June 2008)

- Splicing removes introns (unexpressed regions) and joins exons (expressed regions) to form functional mRNA.
 - Splicing requires a spliceosome formed of special proteins and small nuclear RNAs (snRNAs). snRNAs usually consist of 100 nucleotides. The base composition of snRNA is complementary to the ends of introns.
 - This helps snRNA and introns to base pair and this leads to loop formation. Finally, removal of intron loop and joining of exons takes place.
 - A disease related to this process is: **Systemic Lupus erythematosus (SLE)**: is an autoimmune disease characterized by autoantibodies against many cellular components.
 - Small nuclear ribonucleoproteins (snRNAs) are one of the targets of these antibodies.
 - antibodies.
-

2. The main steps of DNA repair. (September 2009)

1-Recognition of the lesion: And this is done by an endonuclease, which cleaves the damaged strand to form a cut.

2-Excision of damaged DNA: Damaged part is removed by excision exonuclease.

3-Filling the gap: Catalyzed by DNA repair polymerase.

4-Ligation: Catalyzed by DNA ligase.

3. Mechanism of synthesis of mRNA in eukaryotes. (2010)

1-Template binding and formation of Preinitiation Complex (PIC):

-This step requires general transcription factors in addition to pol II.

-General transcription factors (**GTFs**) (TFIIA, TFIIB, TFIID, TFIIE, TFIIF and TFIIH) **promote** RNA polymerase II to transcribe essentially all genes.

-**TFIID which** binds to the TATA box promoter element is the only of these factors that is independently capable of specific high affinity binding to promoter DNA.

(TFIID consists of TATA binding protein (TBP) and 14 TBP-associated factors (TAFs)).

-Binding of the other transcription factors, including TFIIF, which brings RNAP with it.

2-Initiation:

-TFIIH (which has a helicase activity) separates the two strands of DNA

-Phosphorylation by a specific kinase produces activation of the polymerase II.

-Release of TFII A, B, E & H. Then, Pol II-TFIIF complex leaves the promoter region, moves towards the +1 nucleotide and starts to synthesize a complementary transcript of the template DNA strand.

3-Elongation: (This step continues until a termination signal is reached)

-RNA complex utilizes ribonucleoside triphosphates (ATP, GTP, CTP and UTP) and releases pyrophosphate each time a nucleotide is added to the chain.

4-Termination:

-Transcription proceeds till the termination consensus sequence AAUAAA is reached.

-After RNA II has traversed the region of the transcription, RNA polymerase II is dephosphorylated and thus inactivated by a phosphatase enzyme.

-RNA endonucleases cleave the primary mRNA transcript. Polymerase II-TFIIF complex is dissociated. A new cycle of transcription may start again

4. Post-transcriptional modifications for mRNA and the importance for each modification. (2010/2015)

5. Post transcriptional modifications of messenger RNA. (2014)

6. Three types of post transcriptional modifications of mRNA.

(2013) q1-Capping at 5'end: Methyl-guanosine cap is added at the 5' end through:

-RNA triphosphatase hydrolyzes the terminal Y-phosphate of nascent premRNA to form diphosphate.

-The diphosphate is then capped with GMP by the enzyme RNA guanylyl transferase.

-GMP is methylated at position 7 by the use of S-adenosyl-methionine.

- **Importance:**

- a) It protects the 5' end from ribonucleases which are specific for 3'-5' phospho di-ester bonds.
- b) It facilitates protein synthesis through binding with a specific cap binding proteins (initiation factor-4 or IF-4).

2-Polyadenylation at 3' end:

-Poly (A) polymerase (PAP) is responsible for addition of poly (A) tail at the 3' end of pre-mRNA.

- **Importance:**

- a) It stabilizes the mRNA and protects it against the attack by ribonucleases. The length of poly (A) tail determines the half life time of mRNA.
- b) It binds poly A tail-binding proteins which increases the efficiency of translation.

3-Splicing:

-Parts of mRNA do not contain any genetic information.

-These unwanted or intervening sequences are known as introns. Parts of mRNA which carry genetic information are called exons.

-Splicing removes introns (non-expressed regions) and joins exons (expressed regions) to form functional mRNA.

-Splicing requires a spliceosome formed of special proteins and small nuclear RNAs (snRNAs). Usually snRNAs consist of about 100 nucleotides. The composition of snRNAs is complementary to the ends of introns, this helps snRNAs and introns to base pair and this leads to loop formation.

-Adjacent exons come together.

-Removal of intron loop and joining of exons takes place.

- **Importance:**

- a) It decreases the incidence of mutations.
- b) Alternative splicing may lead to the formation of different types of new types of mRNA molecules or proteins.

4-mRNA editing:

Coding information of mRNA can be changed by RNA editing.

In the liver: The single Apo lipoprotein gene (Apo B gene) is transcribed into an mRNA that directs the synthesis of apoB-100 protein (100 kDa)

In the intestine : the same gene directs the synthesis of a primary transcript, then by the action of cytidine deaminase a CAA codon in mRNA is converted to UAA which is a termination codon.

Apo B-48 protein (48 kDa) is the result of translating this edited form of mRNA.

Apo B-100 and Apo B-48 proteins have different functions.

Give an account:-

1 -Base pairing rule in DNA. (2001)

In DNA secondary structure (formation of the double helix),

- The two strands are held together by the complementary base pairing through specific hydrogen bonds.
- Adenine pairs with thymine through 2 hydrogen bonds, and guanine pairs with cytosine through 3 hydrogen bonds.
- Therefore the number of Adenine bases = the number of Thymine bases and the number of Guanine bases = the number of Cytosine bases in DNA.
- Accordingly, the sequence of the two strands is complementary & the sequence of one strand determines the sequence of the second one. This is important during DNA replication as each of the original DNA strands acts as a template for synthesis of a new complementary strand to form 2 daughter DNA molecules.

2 -Mitochondrial DNA. (2004/2011)

- * Mitochondrial DNA forms 0.3 to 1% of the total cellular DNA.
- * Human mtDNA is present in the form of small double stranded circular supercoil (16,569 base pairs 'bp').
- * It's responsible for controlling synthesis of: 2 rRNA, 22 tRNA & 13 proteins, most of these mitochondrial proteins are involved in oxidative phosphorylation (electron transport chain & ATP synthesis).
- * mtDNA molecules are maternally inherited & are used as maternal lineage.
- * Mutations of mtDNA are associated with certain types of myopathies.

3 -Cell cycle restriction point and check points. (2004)

***Cell cycle restriction point:**

- In the cell cycle, there is a restriction point (in late G1) beyond which the cells become committed to enter the S phase & to complete the cycle independent on the presence of the growth factors.*(If cultured mammalian cells are removed from*

medium containing growth factors to one lacking growth factors before they have passed the restriction point, the cell don't enter the S-phase).

***Cell Cycle Check Points:**

- There are 3 check points to ensure that DNA & chromosomal structure are intact before the cell cycle is completed.

1)G1 checkpoint	Check for cell size, nutrients, growth factors & DNA damage.
2)G2 checkpoint	Check for cell size & DNA damage.
3)Spindle assembly checkpoint	Check for proper attachment of chromosomes to spindle.

4 -Degeneracy of genetic codons and wobble theory. (2005)

*** Degeneracy of genetic codons:**

- It's the existence of more than one code word for an amino acid. A given codon codes only one amino acid. But an amino acid may be indicated or coded by more than one codon known as synonym codons. (e.g Arginine is coded by 6 synonym codons).

*** Wobble theory:**

- The mechanism by which tRNAs can recognize more than one codon for a specific amino acid is described by "***The wobble hypothesis***" in which only first two nucleotides are essential, the 3rd nucleotide is flexible, i.e., even if 3rd nucleotide is changed in a codon, that codon may indicate the same amino acid.(***The base pairing of 3rd nucleotide of codon with the 1st nucleotide of anticodon is less specific***)

-The wobble hypothesis is explained by:

- 1) The unusual base pairs can form *because of* flexibility of the bases in the anticodon loop of the tRNA.
- 2) The pairing of the 1st base of the anti-codon with the 3rd base of the codon follows less rigid requirements than the other two.

► The rules that control base pairing of the anticodon first position with the codon third position is indicate in the following table:

Codon 3 position	Anticodon 1 position	rd	st
G	C		
U	A		
A or G	U		
U or C	G		
U, C or A	I		5- DNA

supercoiling. (2005)

- The packing of large DNA molecules into cells requires a process called "DNA supercoiling".
- Supercoiling may be positive (more tight) or negative (less tight). "negative supercoils are more commonly present under normal physiological state".

* Types of supercoiling in linear DNA:

- Toroidal:** The DNA may coil into a series of spirals around cylinder-shaped object.
- Interwound coil:** The DNA may also cross over and under itself repeatedly.

* Types of supercoiling in circular DNA:

- Right-handed supercoil.
- Left-handed supercoil.

6 -Levels of supercoiling of DNA chromatid. (September 2009)

1-The first level of supercoiling: (*packing ratio of 10*)

around histone octomer in the form of nucleosomes produces a ten fold shortening of the length of DNA to form the **10-nm fibril** (10 nm in diameter).

2-The second level of supercoiling: (*packing ratio of 50*)

Requires the presence of H1, this leads to 50-fold shortening of the DNA & it looks like a *solenoid* (cylindrical coil), each turn contains 6 nucleosomes that form the **30nm fiber** (30 nm in diameter).

- * phosphorylation of H1 correlates with chromosomal condensation, due to modulation of the affinity between H1 & histone octomer. **3-The third level of supercoiling:** (*packing ratio of 8000*)

Is the coiling of the 30-nm fiber into twisted-looped structure attached to a protein scaffold in the form of **rosettes**. Each rosettes contains **6 loops**.

Additional levels of supercoiling lead to chromosomal structure & increase the packing ratio to about 8000.

7 -Types and functions of RNA polymerases in prokaryotes and eukaryotes. (2010)

1)Types & functions of RNA polymerases in prokaryotes:

* RNA polymerase "DNA dependent RNA polymerase": Holoenzyme consists mainly of sigma factor(ζ) & Core enzyme: **a) Sigma factor:**

-Enables RNA polymerase to recognize the specific promoter regions in the DNA.

-Increases the affinity of the holoenzyme to the promoter regions. **b)**

core enzyme :

"5 subunits" (1ω , 2α , 1β , & $1\beta'$) which are responsible for the $5'\rightarrow 3'$ RNA polymerase activity. *(this enzyme lacks specificity, i.e. can't recognize the promoter region on the DNA template).*

-Its function: Transcription in prokaryotes.

2)Types & functions of RNA polymerases in eukaryotes:

* **RNA polymerase I:** for synthesis of 45S rRNA, which is cleaved to release the mature 18S, 5.8S & 28S rRNA.

* **RNA polymerase II:** for synthesis of mRNAs, most small nuclear RNAs (snRNAs) & micro RNAs (miRNAs).

* **RNA polymerase III:** for synthesis of tRNAs, 5S rRNA & one snRNA.

(Binds to the promoter and reads the template DNA from $3'\rightarrow 5'$ & synthesizes RNA from $5'\rightarrow 3'$).

8 -The characteristics of genetic code.(five are required)

(2014)

1) Specificity: Each amino acid is coded by a specific sequence of 3 nucleotides, known as codon.

- One codon acts as **initiation** codon for protein synthesis i.e. **AUG**.

- Three codons that don't code for any amino acid called **nonsense** or **termination** codons i.e. **UAA**, **UAG** or **UGA**.

2) Degeneracy of genetic code: It's the existence of more than one code word for an amino acid. A given codon codes only one amino acid. But an amino acid may be indicated or coded by more than one codon known as synonym codons. *(e.g Arginine is coded by 6 synonym codons).*

3) Universality: The genetic code is mainly universal with the exception of few examples e.g. mitochondrial RNA reads 4 codons differently from the cytoplasmic RNA.

4) Reading frame: Usually one reading frame will produce a functional protein *(it's determined by the initiation code)*. The correct reading frame is set in vivo by

recognition of the initiation codon (AUG) by the ribosomes at the start of the coding sequence.

5)Wobble: The mechanism by which tRNAs can recognize more than one codon for a specific amino acid is described by "**The wobble hypothesis**" in which only first two nucleotides are essential, the 3rd nucleotide is flexible, i.e., even if 3rd nucleotide is changed in a codon, that codon may indicate the same amino acid. (**The base pairing of 3rd nucleotide of codon with the 1st nucleotide of anticodon is less specific**).

- **The wobble hypothesis is explained by:**

- 1)The unusual base pairs can form *because of* flexibility of the bases in the anticodon loop of the tRNA.
- 2)The pairing of the 1st base of the anti-codon with the 3rd base of the codon follows less rigid requirements than the other 2.

9 -Lactose Operon.

(2014) *Components and mechanism of action in presence of lactose & absence of glucose (2015)

-Lactose operon of E-coli is one of the best-understood examples of the operon, which is concerned with the metabolism of lactose. The lac operon allows for effective metabolism of lactose when glucose isn't available.

-The components of lactose (Lac) Operon:

A) Structural genes (inducible):

1-**The lacZ gene:** codes for **β-galactosidase**, which hydrolyzes lactose to galactose & glucose.

2-**The lacY gene:** codes for a **permease** that facilitates the movement of lactose into the cell.

3-**The lacA gene:** codes for **thiogalactoside transacetylase** whose exact physiologic function is unknown. **B)Regulatory genes:**

1-**Regulatory lac I gene:** it's constitutive gene. It encodes a repressor protein.

2-**Promoter:** where RNA polymerase binds.

3-**Operator (O) site:** where the repressor binds.

4-**Catabolite gene activator protein (CAP) binding site:** where a regulatory protein termed catabolite gene activator protein binds.

- Mechanism of action of lac operon in presence of lactose & absence of glucose:

The lac operon is induced (turned on).

*A small amount of lactose is converted to an isomer.

* This isomer is an inducer that binds to the repressor protein, changing its conformation so that it can no longer bind to the operator.

- * In the absence of glucose, adenylyl cyclase is active, & sufficient quantities of cAMP are made & bind to the CAP protein.
- * The cAMP-CAP complex binds to the CAP-binding site, causing RNA polymerase to more efficiently initiate transcription at the promoter site.
- * This is an example of positive regulation.
- * The transcript is a single polycistronic mRNA molecule that contains 3 sets of initiation & stop codons. (*Translation of the mRNA produces the 3 proteins that allow lactose to be used for energy production by the cell*).

10-DNA secondary structure. (2013/2014)

Watson & Crick proposed a structure for DNA in the form of a double helix & it's now referred as B-form of DNA, which is the most common physiological form. It has the following characters:

1) Two antiparallel strands from a right-handed helix:

- The 2 strands of DNA are paired to each other & coil around a common axis to form a right-handed helix.
- The 2 strands run antiparallel, one runs in the 5' to 3' direction & the other in the 3' to 5' direction.

2) Complementary base pairing:

- The two strands are held together by the complementary base pairing through specific hydrogen bonds.
- Adenine pairs with thymine through 2 hydrogen bonds, and guanine pairs with cytosine through 3 hydrogen bonds.
- Therefore the number of Adenine bases = the number of Thymine bases and the number of Guanine bases = the number of Cytosine bases in DNA.
- Accordingly, the sequence of the two strands is complementary & the sequence of one strand determines the sequence of the 2nd one. This is important during DNA replication as each of the original DNA strands acts as a template for synthesis of a new complementary strand to form 2 daughter DNA molecules.

3) Base stacking:

- The base pairs inside the helix are stacked above each other by ***Van der Waals forces & hydrophobic interactions***.
- The ***hydrogen bonding*** between complementary base pairs & the ***Van der Waals forces & hydrophobic interactions*** of stacked base pairs provide the stability of the double helix.
- The excessive stacking of DNA is balanced by ***the repulsive forces*** of negative charges present on phosphate groups.

4) Spiral staircase:

-The double helix of DNA appears much like spiral staircase, in which there are: 10.4 base pairs or steps for each complete turn of the helix.

-Each complete turn is 3.4 nm long.

5) **Dimensions:**

-The B-form is 2 nm wide.

-From outside of the helix, 2 grooves are apparent:

*a major groove (2.2nm).

*a minor groove (1.2nm).

Through these grooves many drugs and proteins can make contact with the nitrogenous bases without any need to open the helix.

11- DNA primary structure. (2014)

* The nucleotides that form DNA primary structure are mainly 4: dAMP, dGMP, dCMP & dTMP.

* In each strand, the nucleotides are linked together by phosphodiester bonds between the 5'-hydroxyl of one nucleotide & 3'-hydroxyl of the next one.

* The alternating sugar phosphate units form the backbone of each DNA strand (5'P-S-P-S-3').

* The nitrogenous bases, which are linked to the pentoses, are projecting to the inside of the 2 strands of DNA at right angle.

* The sequence of bases determines the coding structure of DNA (genetic information).

* Each polynucleotide strand has 2 terminals, One end has a free phosphate group attached to 5'-hydroxyl group of the terminal pentose & the other end has a free 3'hydroxyl group.

* The order of nucleotides in any DNA strand is always written in the 5' to 3' direction, which is the direction of synthesis.

Define :

RNA splicing:

- It is the removal of introns (non expressed regions of RNA) and joining the exons (expressed regions) to form functional mRNA.
- Splicing requires a spliceosome (special proteins and small nuclear RNAs "Usually snRNAs consist of about 100 nucleotides"

Mention :

1.Eukaryotic and prokaryotic replication (4 differences).

Prokaryotic	Eukaryotic
Unique origin of replication. Larger okazaki fragments (1000-2000 nucleotides). Polymerases involved: DNA polymerase III (synthesizes both strands of DNA). DNA polymerase I (removes RNA primers). DNA polymerase II (involved in the proof reading and DNA repair). Topoisomerases: Type I DNA topoisomerase (unwinding the positive supercoils). Type II DNA topoisomerase (separates the interlocked strands).	Multiple origins of replication Small okazaki fragments (100-200 nucleotides) Polymerases involved: DNA polymerase α-primase complex (synthesis of RNA primers & short DNA stretches connected to the RNA primers). DNA polymerase β (DNA repair). DNA polymerase γ (mitochondrial DNA synthesis). DNA polymerase δ (synthesis of the lagging strand). DNA polymerase ϵ (synthesis of the leading strand). Topoisomerases : Type I DNA topoisomerase (removal of positive supercoils in front of the replication fork).

2.DNA and RNA (4 difference).

	DNA	RNA
Sugar	2-deoxyribose.	Ribose.

Site	Nucleus and mitochondria.	Mainly in cytosol, less commonly in nucleus & mitochondria.
N-Bases: a)purines b)pyrimidines	Adenine , Guanine Cytosine , Thymine	Adenine , Guanine Cytosine , Uracil NB:thymine (minor tRNA bases).
Strands	Double helix.	Single strand.
Types	Circular or linear (A,B,Z forms).	mRNA tRNA rRNA
Function	Genetic information and synthesis of RNA.	Protein synthesis.

What is meant by :

☒ **DNA-dependant-RNA polymerase: (June 2008).**

*An enzyme responsible for initiation of DNA replication in prokaryotes as it forms RNA primers to be elongated by DNA polymerase III, it is also known as **primase**.

*It can be defined as: the major enzyme of transcription it catalyzes the synthesis of all 3 types of RNAs like (mRNA, tRNA,& rRNA) in prokaryotes like E-Coil, it doesn't require primer to initiate polymerization and doesn't possess nuclease activity

☒ **Promoter : (June 2008)**

The characteristic consensus sequence of nucleotides on the DNA strand it's recognized by RNA polymerases which bind to it to form preinitiation complex to initiate transcription.

In prokaryotes: promoters are TATA box and -35 sequence.

In Eukaryotes: promoters are TATA box and CAAT & GC box.

☒ **Telomerase : (august 2008)**

Enzyme responsible for elongation of 3' end of telomere of DNA lagging strand. In eukaryotes it's a special kind of reverse transcriptase , made of protein part and RNA strand (of about 150 nucleotides long).

☒ **Pribnow or TATA box :**

It's a promoter in prokaryotes formed of six nucleotides

"5' TATAAA 3' " centered about 10 nucleotides to the left the transcription start site.

Give an account on :

1) **Point mutation (2004).**

Base substitution (point mutations):

- They are the most common type and include two subgroups:

Transition	Transversion
In which one purine is replaced by another purine or one pyrimidine is replaced by another pyrimidine.	In which a purine is replaced by pyrimidine or a pyrimidine is replaced by purine .

Base substitutions may occur in the coding region of gene , and can produce one of the following effects : **missense** , **nonsense** or **silent mutation**.

a) **Missense mutation:**

Leads to a changed codon and results in an amino acid change in the protein product of that gene. (*Depending on the amino acid changed , missense mutations can have either no effect or very serious changes e.g. Sickle cell anemia*).

b) **Nonsense mutation :**

Leads to conversion of an amino acid codon to a stop or nonsense codon. (*Translation of mRNA from that gene is prematurely terminated and the protein product s usually non- functional e.g. thalassemias*).

c) **Silent mutation :**

Leads to formation of a synonym codon that codes for the same amino acid and no change occurs in the amino acid sequence of the gene products.

Compare:

The effect of missense mutation and silent mutation (2015).

Missense mutation	Silent mutation
Leads to a <u>changed codon</u> and results in <u>an amino acid change</u> in the protein product of that gene. (<i>Depending on the amino acid changed , missense mutations can have either no effect or very serious changes e.g. <u>Sickle cell anemia</u></i>).	Leads to formation of a <u>synonym codon</u> that codes for the same amino acid & no change occurs in the amino acid sequence of the gene products.

Describe the following processes :

1. Cis-acting regulatory mechanisms of gene expression (June 2008):

- Cis-acting elements are DNA , regulatory sequences flanking a gene.
- They are usually embedded in the non-coding regions of the genome.
- They influence expression of genes only on the same chromosome.
- The interaction between these DNA segments and regulatory molecules , such as transcription factors , can induce or repress the transcriptional machinery , influencing the kinds and amounts of products that are produced.

2. Types of environmentally induced DNA errors (mutations). (August 2008). I.

Base substitutions (point mutations):

1- Transition. 2-

Transversion:

- a) Missense mutation.
- b) Nonsense mutation. C) Silent mutation. I.

Deletion of one or more bases:

- a) Frame shift mutation
- b) Deletion of three or multiple of three bases I.

Insertion of one or more bases:

- a) Frame shift mutation.
- b) Deletion of three or multiple of three bases.

3. the importance of capping and splicing of mRNA:

*Importance of capping:

- It Protects the 5'-end from ribonucleases which are specific for 3'5' phosphodiester bonds.
- It facilitates protein synthesis through binding with a specific cap binding proteins (initiation factor-4 or IF-4).

*Importance of splicing:

- It decreases the incidence of mutations.
- Alternative splicing may lead to the formation of different types or new types of mRNA molecules or proteins.

4. The role of sigma subunit in initiation of transcription. (2014).

- Enables RNA polymerase to recognize the specific promoter region on the DNA.
- Increases the affinity of the holoenzyme to the promoter region.

5. The mitochondrial pathway of apoptosis. (2014)

- It's used in response to cellular stress (DNA damage, growth factor withdrawal, exposure to cytotoxic drugs, cell cycle perturbation).
- These stressors promote release of cytochrome C from mitochondria.
- Once released into cytosol, cytochrome c associates with the apoptotic proteaseactivating factor1 (Apaf 1) and procaspase-9 to form **apoptosome**, which activates caspases 9, leading to activation of an **effector caspase**, then activation of **CADase** and induction of apoptotic process.

Enumerate

1. Two minor bases, one purine & one pyrimidine present in tRNA. (2000).

*Minor pyrimidine is: Dihydrouracil.

*Minor purine is: 7-methylguanine or N⁶ methyladenine or N⁶,N⁶ dimethyladenine.

2.

Two DNA helical forms , one is broad cast and one is left handed. (2000).

*Broad cast(B-form DNA). *left handed (Z form DNA).

3. - four pyrimidine bases found in nucleic acids. (sep 2009).

* Cytosine.

*Uracil.

*Thymine.

*Dihydrouracil.

*5-Methylcytosine.

*5-Hydroxymethylcytosine.

4. **four adenine containing nucleotides.** *Adenosine monophosphate (AMP).
*Adenosine diphosphate (ADP).

*Adenosine triphosphate (ATP)

*Cyclic 3',5' Adenosine monophosphate (cAMP).

*S – adenosyl methionine (SAM).

*3' phosphoadenosine-5'-phosphosulfate (PAPS).

*Nicotinamide Adenine Dinucleotide (NAD⁺).

*Nicotinamide Adenine Dinucleotide phosphate (NAP⁺).

5. **- Four characteristics of genetic code.**

1- **specificity.**

2- **degeneracy of genetic code.**

3- **universality.**

4- **reading frame.**

6. **Three examples of tumor markers and mention the importance of each one (August 2008).**

- **Prostate specific antigen (PSA)** for prostate cancer .
- **Human chorionic gonadotropin (HCG)** for gestational trophoblastic tumors and some germ cell cancers .
- **Alpha fetoprotein (AFP)** for certain germ cell cancers and liver cancer .
- **Carcinoembryonic antigen (CEA)** for colon and rectal cancers .
- **Cancer antigen (CA 125)** for ovarian cancer .
- **CA 15-3** for breast cancer .
- **CA 19-9** for colorectal and pancreatic cancers .

7. **Four mechanisms for conversion proto-oncogenes into oncogenes . (2009/2012).**

- **Promoter or enhancer insertion** : many viruses act through this mechanism.
 - **Gene amplification** : it is present in many tumor cells .
 - **Point mutation** : this produces changes in the protein product of the gene .
 - **Chromosomal translocation** : *For example* , Chromosomal translocation between chromosome 9 and 22 results in formation of **PHILADELPHIA CHROMOSOME** that produces increased activity of the tyrosine kinase and transforms normal cells to leukemic cells .
-

8. Four causes of gene mutation :(2009/2013).

- *Errors in replication.
- * Errors due to recombination events.
- *Chemical mutagens.
- *Irradiation.
- * oxidative damage of DNA.

**9. Four post translational covalent alteration of proteins (2009/2012).
two types of post translational covalent modification (2014).**

- Phosphorylation. - Glycosylation.
- Acetylation or methylation.
- Hydroxylation.
- carboxylations.

☒ **Mention the differences between: the effect of missense and nonsense mutations.**

.) Both are effects of base substitutions (point mutation	
Nonsense mutation	Missense mutation
Conversion of an amino acid codon to a stop or nonsense codon.	Change in the codon.
Translation of the mRNA from that gene is terminated prematurely and then the protein product is nonfunctional.	Results in an amino acid change in the protein product of that gene.
Can leads to (Thalassemias).	Depending on the amino acid changed. It can have no effect or serious change. (Sickle cell anemia).

Explain on biochemical basis :

1) DNA damage may lead to apoptosis (2007)

(The mitochondrial pathway of apoptosis "intrinsic pathway ").

- It's used extensively in response to diverse forms of cellular stress (DNA damage, growth factor withdrawal, cell-cycle perturbation, exposure to cytotoxic drugs).
- It is considered as a form of cellular stress which promote release of **cytochrome C** from mitochondria.

- Once released into the cytosol, Cytochrome c associates with the apoptotic protease activating factor 1 (Apaf1) and procaspase-9 to form apoptosome, which activates caspase 9, leading to activation of an effector caspase, then activation of CADase and induction of apoptotic process.

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2) Tumor markers are used to monitor cancer except if cancer is very sensitive to chemotherapy. (2007).

(Evaluation of effectiveness of treatment)

Certain markers found on cancer cells can be used to help predicting if a certain treatment is likely to work or not. For example, in breast cancer, finding estrogen receptors in breast tissue is used to detect good response to anti-estrogen therapy.

3) Explain how tumor markers are used for diagnosis, follow up and screening of tumors.

Illustrate your answer with examples. (June 2008)

- Screening and early detection of cancer: a perfect tumor marker could be used as a cancer screening blood test for all people. The most widely used tumor marker is the prostatic specific antigen (**PSA**) blood test. The PSA is used to screen for prostatic cancer. Tumor markers are usually not used to diagnose cancer. In most cases, cancer can only be diagnosed by a biopsy.
 - Determining the prognosis for certain cancers: Some types of cancer grow and spread faster than others. This may even occur within the same cancer type. For example, in testicular cancer, very high levels of tumor marker like human chorionic gonadotropin (**HCG**) or alpha fetoprotein (**AFP**) predicts a more aggressive cancer. Patients with these high levels may be given more aggressive treatment. [Explain:](#)
- 1. p53 is called the molecular policeman. (2011-12-13-2015).**
- When the DNA damage is **too severe** to allow effective repair, **p53** acts through **activation of Bax gene** and triggers apoptosis. So, this will lead to the death of the cell, That's why **p53** is called the **molecular policeman** or **Guardian of the genome**.

2. For protein synthesis, 20 amino acids are utilized, They are capable to charge about 50 tRNA species.

3. A charged tRNA may recognize more than one codon. using translation process. (2007)

(Wobble hypothesis).

-a tRNAs can recognize more than one codon for a specific amino acid it is described by "**The wobble hypothesis**" in which only first two nucleotides are essential, the 3rd nucleotide is flexible, i.e., even if 3rd nucleotide is changed in

a codon, that codon may indicate the same amino acid. (*The base pairing of 3rd nucleotide of codon with the 1st nucleotide of anti-codon is less specific*).

4. One cannot predict the sequence of nucleotides in gene knowing amino acid sequence in the protein coded the gene. (2010/11/12/13).

- Because it may be more than one code word for an amino acid. A given codon codes only one amino acid. But an amino acid may be indicated or coded by more than one codon known as **synonym codons**. For example, arginine is coded by 6 synonym codons.

5. Bases substitution may have effects or may cause serious effects. (2014). - Bases substitution may occur in coding region of a gene and produce:

1) Missense mutation:

Leads to a changed codon and results in an amino acid change in the protein product of that gene. (*Depending on the amino acid changed , missense mutations can have either no effect or very serious changes e.g. Sickle cell anemia*).

2) Nonsense mutation :

Leads to conversion of an amino acid codon to a stop or nonsense codon. (*Translation of mRNA from that gene is prematurely terminated and the protein product s usually non- functional e.g. thalassemias*).

3) Silent mutation :

Leads to formation of a synonym codon that codes for the same amino acid and no change occurs in the amino acid sequence of the gene products.

6. Sickle cell anemia is a genetic disorder caused by point mutation (2014).

- Sickle cell anemia occurs due to, missense mutation (a change in the codon that results in an amino acid change in the protein product of the gene), & missense mutation is an effect of point mutation.

